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Effect of ripeness and postharvest storage on the evolution of colour and anthocyanins in cherries (*Prunus avium* L.)

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Abstract

The relationship between colour parameters and anthocyanins of four sweet cherry cultivars, Burlat, Saco, Summit and Van was studied. The colour (L^*, a^*, b^*, chroma and hue angle parameters) and anthocyanins were analysed during two different years at two different ripening stages (partially ripe, and ripe, respectively). The cherries were analysed at harvest and after storage at 1.5 ± 0.5 °C and 15 ± 5 °C for 30 and 6 days, respectively. The colour was measured by tristimulus colourimetry (CIELAB system) directly on the fruits, while anthocyanins were quantified by HPLC-DAD analysis on methanolic extracts of freeze-dried samples of the fresh cherries and on the differently stored cherries. L^*, chroma, and hue angle values were always lower for the ripe than for the partially ripe cherries. All of the cultivars were found to contain cyanidin-3-rutinoside and cyanidin-3-glucoside as the major anthocyanins. The total anthocyanin content in fruits of the different cultivars varied in the order Burlat > Saco > Van > Summit. The concentration of anthocyanins increased at both temperatures of storage in both ripe and partially ripe cherries, but the extent of increase varied among cultivars. Cherries stored at 15 ± 5 °C showed higher reduction of L^*, chroma and hue angle than fruits stored at 1.5 ± 0.5 °C. L^*, a^*, b^*, chroma and hue angle correlated negatively ($P < 0.001$) with the total anthocyanins levels, but not with the total phenols. These results show that chromatic functions of chroma and hue correlate closely with the evolution of colour and anthocyanins levels during storage of sweet cherries and indicate that colour measurements can be used to monitor pigment evolution and anthocyanin contents of cherries (and *vice versa*).

Keywords: Anthocyanins; Cherries; Chromatic coordinates; Chroma; Colour evolution; Hue angle; Ripeness stage; Storage

1. Introduction

Colour is one of the most important indicators of maturity and quality of fresh, stored, and processed cherries (Drake, Proebsting, & Spayd, 1982). In cherries, colour is mainly influenced by the concentration and distribution of different anthocyanins in the skin (Gao & Mazza, 1995) as well as pH and levels and types of colourless phenolics in the fruits and other factors such as light, temperature, oxygen, metal ions and enzymes (Delgado-Vargas & Paredes-López, 2003).

In a previous study we showed that levels of anthocyanins in four sweet cherry cultivars, Burlat, Saco, Summit and Van, ranged from ~5 to 86 mg/100 g of fresh weight (fw) in 2001, and from ~6 to 230 mg/100 g of fw in 2002 (Gonçalves et al., 2004). In general, the total anthocyanins levels were higher in 2002 than in 2001, but the profiles of anthocyanins were similar both among the two years and among all four cultivars (Gonçalves et al., 2004). The

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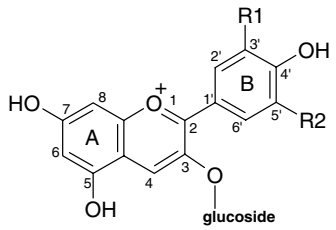

R_1	R_2	Anthocyanin	Glucoside	Colour
H	H	Pelargonidin	rutinoside	Red
OH	H	Cyanidin	glucoside	Orange-red
OH	H	Cyanidin	rutinoside	Red-purple
OCH_3	H	Peonidin	glucoside	Orange-red
OCH_3	H	Peonidin	rutinoside	Orange-red

Fig. 1. The basic chemical structure of the six most commonly occurring anthocyanins in sweet cherries. The structures shown the hydroxylation and methoxylation substitution pattern, type of glycosidic residue, and colour of the flavylium ion form (prevailing at acidic pH).

major anthocyanins in sweet cherries include the 3-*O*-glucoside and 3-*O*-rutinoside (-rhamnosyl-D-glucopyranose) of cyanidin, with peonidin-3-*O*-rutinoside and -glucoside, as well as pelargonidin-3-*O*-rutinoside occurring in much lower amounts (Fig. 1) (Esti, Cinquanta, Sinesio, Moneta, & Di Matteo, 2002; Gao & Mazza, 1995; Gonçalves et al., 2004). Gao and Mazza (1995) reported that the total anthocyanin content ranged from 82 to 297 mg/100 g for dark cherries and from 2 to 41 mg/100 g for the light coloured cherries. The cyanidin-3-rutinoside and the cyanidin-3-glucoside contents in pitted, sweet cherry cultivars have been found to range from 4 to 44 mg/100 g of fw and from 2 to 243 mg/100 g of fw, respectively (Gao & Mazza, 1995).

As expected, the total levels of anthocyanins are higher in ripe cherries than in partially ripe ones (Gonçalves et al., 2004). In freshly harvested, fully ripe cherries, the levels of cyanidin-3-rutinoside were in our previous study found to represent 63–94% by weight of the total anthocyanins. In both years, cherries of cv. Burlat showed the highest anthocyanin levels, particularly with respect to the cyanidin-3-glucoside levels. The levels of anthocyanins increased during storage, and after storage the total anthocyanins represented around 50% of total phenolics. The cherries from cv. Van exhibited the most profound increase in total anthocyanin levels during storage, and the increase in total anthocyanin levels was mainly attributable to increases in the cyanidin-3-rutinoside level (Gonçalves et al., 2004). Studies for anthocyanin levels in mature grapes stored at 0 °C supported our findings, since a slight increase, from 4500 to 6000 mg/kg of peel fw, was shown previously for these compounds (Cantos, García-Viguera, Pascual-Teresa, & Tomás-Barberán, 2000).

The colorimetric CIE system, Commission International d'Eclairage, is widely used in the quantification and characterization of anthocyanin chromatic properties and in the assessment of colour quality and colour changes during maturity and processing of plant foods (Dodds, Brown, & Ludford, 1991; Heredia, Francia-Aricha, Rivas-Gonzalo, Vicario, & Santos-Buelga, 1998).

Fresh sweet cherries represent an important, but fragile, commodity in the Portuguese agricultural export market. The harvesting season is very short, and cold storage is used to stretch the supply period in the season. However, the effects of different storage conditions on cherry quality, including colour development, is not well studied and the available knowledge on anthocyanins levels versus colour development during cherry storage appears somewhat confusing. In a study of two sweet Italian cherry cultivars, Sciazza and Ferrovia, it was found that the content of cyanidin-3-rutinoside and cyanidin-3-glucoside in the cherries decreased several fold during cold storage for 15 days at 1 °C (Esti et al., 2002). Nevertheless, the colour attributes of the same cherries, measured as CIE: L^*, a^*, and b^* values of the cherry skins and pulp, did not change significantly during storage independent of the storage temperature – the only exception being a decrease in L^* observed for the skin of the cv. Ferrovia cherries (Esti et al., 2002). In contrast, as discussed above, we previously observed increases in anthocyanin levels during storage suggesting that colour attributes would change during storage (Gonçalves et al., 2004). This study was therefore undertaken to: (a) determine the evolution of colour in sweet cherries during postharvest storage at different temperatures, (b) assess the relationship between colour attributes and anthocyanins content in cherries, and (c) unravel any potential differences in colour attributes and anthocyanin-colour relationships among four different cherry cultivars.

2. Materials and methods

2.1. Cherry raw material

Four sweet cherry cultivars, Burlat, Saco, Summit and Van from an orchard in Vila Real, Portugal, were randomly hand harvested in 2001 and 2002, both years as partially ripe and ripe. The fruit analyses were made at harvest and during storage at typical, industrially used fruit storage conditions, that is at 1.5 ± 0.5 °C and 90% RH (cool temperature) for 30 days or at 15 ± 5 °C (room temperature) for 6 days. The details on the quality criteria decisive for picking as well as an elaborate examination of the evolution of quality parameters in the four different cherry cultivars during storage (in addition to effects of maturity stage at picking) have been reported separately (Gonçalves et al., 2004).

2.2. Colour analyses

Ground colour was measured on 20 fruits using a tristimulus colorimeter (Minolta CR-200B Chroma Meter, Minolta, Japan) having an 8 mm diameter viewing area. Chromatic analyses were carried out following the CIE (Commission International de l'Eclairage) system of 1976.

Values of L^*, a^* and b^* were measured to describe a three-dimensional colour space and interpreted as follows: L^* indicates lightness read from 0 (completely opaque or "black") to 100 (completely transparent or "white"). A positive a^* value indicates redness ($-a^*$ is greenness) and a positive b^* value yellowness ($-b^*$ is blueness) on the hue-circle (Hutchings, 1994; Voss, 1992). The hue angle (°), hue = arctg (b^*/a^*), expresses the colour nuance (Voss, 1992) and values are defined as follows: red-purple: 0°, yellow: 90°, bluish-green: 180°, and blue: 270° (McGuire, 1992). The chroma, obtained as $(a^{*2} + b^{*2})^{1/2}$, is measure of chromaticity (C^*), which denotes the purity or saturation of the colour (Voss, 1992). The data of each measurement are the average of triplicate measures on equidistant points of each fruit.

2.3. Chemicals and reagents

Anthocyanin–glucosides were purchased from Polyphenols A/S (Stavanger, Norway) and the HPLC grade acetonitrile was purchased from Merck (Darmstadt, Germany).

2.4. Extraction of anthocyanins

Pitted and freeze-dried cherry samples (0.5 g) were contacted with 60% v/v aqueous MeOH (5 ml); flushed with N_2, and extracted for 10 min using a shaking (200 rpm) water bath at 25 °C. The individual samples were then filtered through one layer of Whatman No. 1 filter paper (using vacuum suction during the filtration) and the solvent contacting was repeated twice on the residue. Each filtrate was then filtered through a 0.45 μm syringe-tip hydrophilic Durapore filter (Millipore Corp., Bedford, MA) prior to high performance liquid chromatography (HPLC) analyses. The reported values are the sums of the values obtained from each extraction and each HPLC analysis.

2.5. Anthocyanins and phenols analyses

HPLC analysis was carried out according to the procedure described by Lamuela-Raventós and Waterhouse (1994) using a Hewlett–Packard 1100 system (Waldbronn, Germany) equipped with a diode array detector (DAD), a Nova-Pak C18 column (3.9 × 150 mm, Waters) at 40 °C, and controlled by a PC with HPChem station Software. The solvent flow rate was 0.5 ml/min and the injected volume was 10 μl. The anthocyanins were identified by spectral and retention time analysis. The quantities of the different phenolic compounds were assessed from peak areas and calculated as equivalents of standard compounds from linear regression curves of authentic standards. The anthocyanins were quantified at 520 nm, in mg/100 g of fw, as cyanidin-3-glucoside and cyanidin-3-rutinoside, respectively, while the quantities of peonidin-3-glucoside, peonidin-3-rutinoside, and pelargonidin-3-rutinoside were calculated as cyanidin-3-rutinoside equivalents.

2.6. Statistics

Analyses of variance were accomplished by use of the Super ANOVA software (1.11 Abacus Concepts Inc., 1991). Significances of differences were established from a Duncan's Test ($P < 0.05$). A Fisher correlation analysis including all the parameters was also performed. Possible differences between cultivars in the correlation between two parameters were analyzed by comparison of regression lines.

3. Results and discussion

3.1. HPLC-DAD analysis of cherry anthocyanins

The HPLC chromatograms of sweet cherry extracts obtained in the visible spectral region (520 nm) revealed five peaks, which corresponded to five anthocyanins (Fig. 2): cyanidin-3-glucoside (peak 1), cyanidin-3-rutinoside (peak 2), peonidin-3-glucoside (peak 3), pelargonidin-3-rutinoside (peak 4) and peonidin-3-rutinoside (peak 5). As discussed below these findings are in complete agreement with what has been reported previously on anthocyanins in sweet cherries (Gao & Mazza, 1995). The details on the evolution of anthocyanins in the four different cherry cultivars during storage (in addition to effects of maturity stage at picking and year) have been reported separately (Gonçalves et al., 2004).

3.2. Effect of ripeness and storage temperature on cherry colour

The chromatic characteristics of the fruits studied are shown in Tables 1 and 2. There were significant differences in L^*, chroma and hue angle ($P < 0.001$) between the four cultivars, ripeness stage, year and storage. At harvest, Burlat cherries always showed lower hue angle and L^* and higher anthocyanin levels in both years, a synonym of redder and darker cherries. Summit fruits showed higher L^* and chroma, which is in accordance with the observation that Summit fruits are both less red and lighter in colour than the other cherry cultivars. L^*, chroma and hue angle of partially ripe cherries were always higher than in the ripe cherries, which means a less red fruit, and was correlated with lower anthocyanin content (Tables 1–3; Gonçalves et al., 2004).

L^*, chroma and hue angle of ripe and partially ripe cherries were always higher in 2001 (Tables 1 and 2). These parameters decreased during storage, mostly at room temperature, but the extent of change varied among the cultivars (Tables 1 and 2). In general, at 15 ± 5 °C, chroma was the parameter that showed higher reduction in 2001 and 2002 (Tables 1 and 2). The decrease in chroma means an increase in the tonality of the fruit colour. A reduction of 19 units was measured in Van cherries in 2001 (Table 1). Rodríguez-Saona, Giusti, and Wrolstad (1999) observed a reduction of 10 units in chroma, in radish-coloured juices

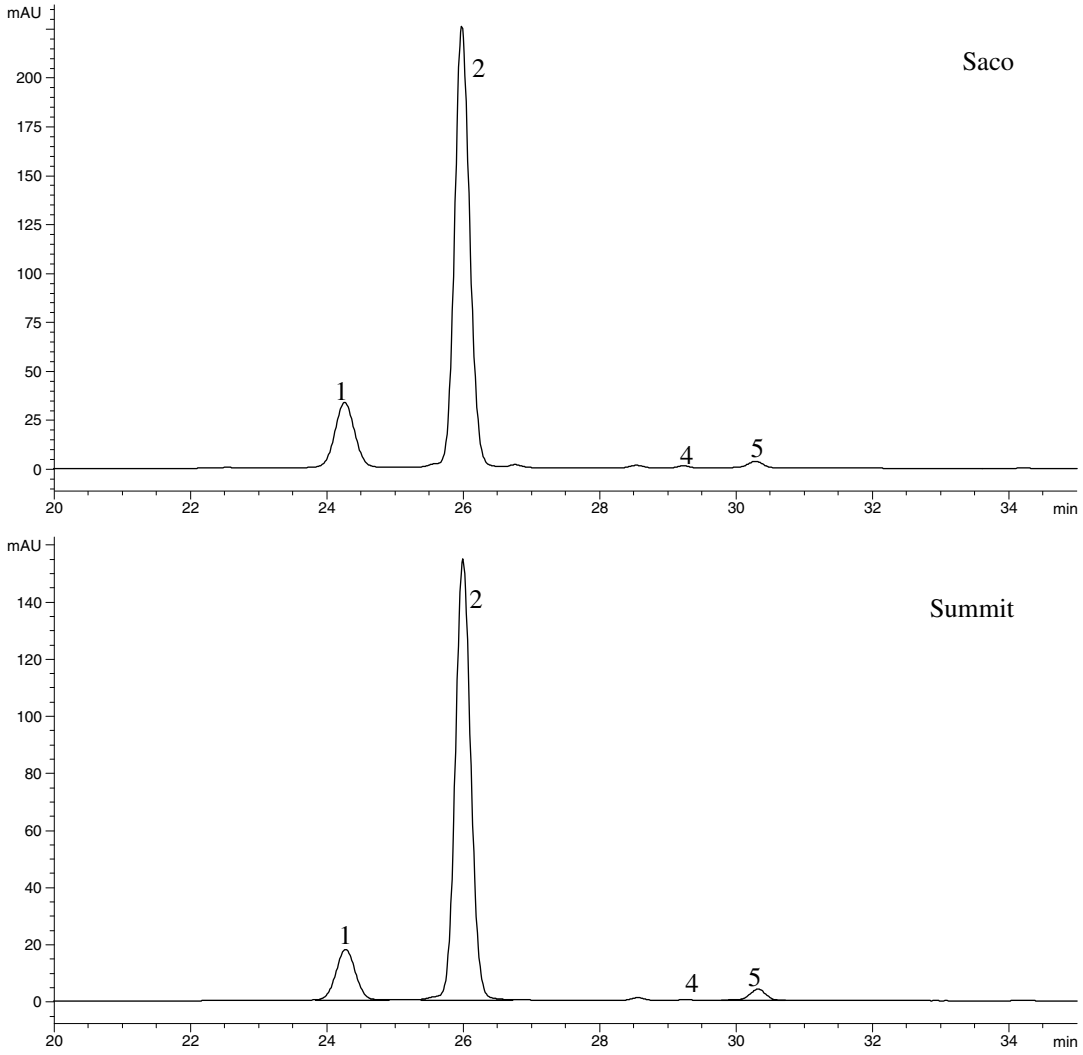

Fig. 2. HPLC separation of anthocyanins in a methanolic extract of the four sweet cherry cultivars, monitored at 520 nm. Peaks: 1, cyanidin-3-glucoside; 2, cyanidin-3-rutinoside; 3, peonidin-3-glucoside; 4, pelargonidin-3-rutinoside; 5, peonidin-3-rutinoside.

after storage at 16 days at 25 °C. A decrease in chroma during fruit storage was also observed with stored strawberries (Abers & Wrolstad, 1979). The decrease could have been caused by an increase in total anthocyanins and a decrease in chlorophyll and carotenoids, and, according to Abers and Wrolstad (1979), by development of dark, pigmented compounds, which tend to mask colour.

All treatments induced a reduction in hue angle values, when compared to the values observed at harvest (Tables 1 and 2). The loss of lightness was reflected by a reduction of L^* (the photometric parameter proportional to the light reflected by the object) and was directly related to the humidity during storage (humidity data not shown). Hence, the cherries of all four cultivars became a little darker as well as a little redder during storage, as shown by decreases in L^* values and in hue angle values (Tables 1 and 2), during both the cool (1.5 °C) and the ambient (15 °C) storage periods. However, the changes in colorimetric parameters varied depending on the storage temperature and the anthocyanins composition in the different cherry cultivars. Refrigerated temperatures greatly improved the colour stability of the cherries. Differences in hue angle could be attributed to both differences in anthocyanin and phenols composition, and to interaction of anthocyanins with other compounds at the relatively low pH of the fruits (intermolecular co-pigmentation). Co-pigmentation is optimal in the range pH 3–5 (Brouillard, Wingand, Dangles, & Cheminal, 1991), and is known to decrease with increasing temperature (Baranac, Petranović, & Dimitrić-Marković, 1996). In addition, the low pH increases the stability of anthocyanins (Eiro & Heinonen, 2002). On this basis we ascribe the fact that cherries can be stored for at least one month at 1.5 °C without degradation of anthocyanins to be due to stabilization by pH and co-pigmentation. However, to firmly establish the mechanism behind the stabilization of the colour of the cherries during storage, further chemical analyses including LC–MS and NMR analyses are warranted.

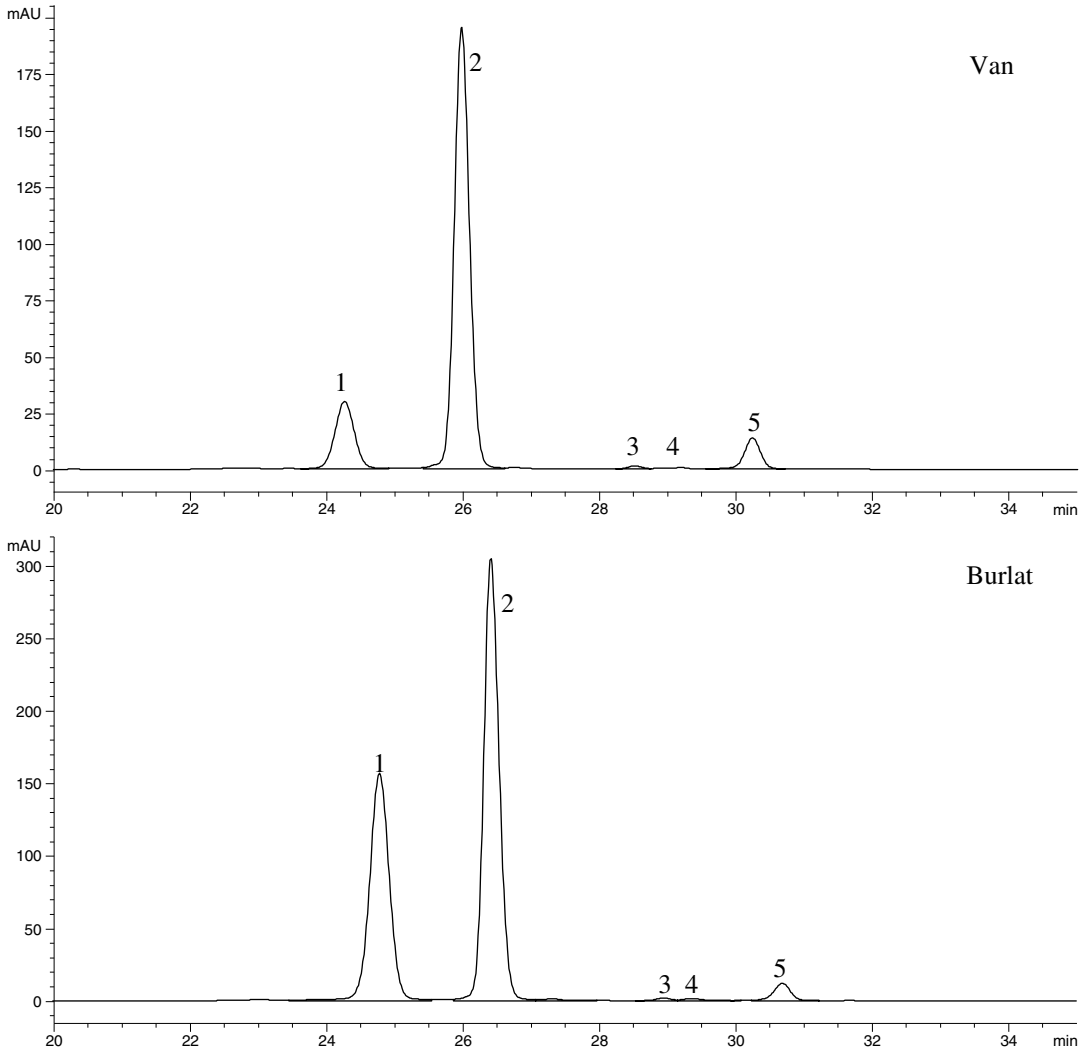

Fig. 2 (*continued*)

3.3. Influence of pH on cherry colour

The decrease in chromatic parameters was associated with a weak increase in the pH (Tables 1 and 2; Gonçalves et al., 2004). The same results were observed by Heredia et al. (1998), who found that the chroma defined in the uniform colour spaces, underwent a linear decrease as pH increased. The influence of pH on fruit colour is well known. As pH increases, the colour of anthocyanins moves to the non-spectral purple and approaches a progressive loss of colour.

3.4. Correlations and regressions between anthocyanins content and colour

With all the cherry cultivars, the levels of anthocyanins correlated negatively with each of the colour parameters L^*, a^*, b^*, chroma and hue angle (Table 3). The steepest and most significant correlations were found for the cultivars having highest levels of anthocyanins, namely Burlat and Van (Table 3). The different correlation coefficients obtained for the different cherry cultivars means that the evolution of colour during storage varied among the different cherry cultivars. Some of the main aims of the present work were to assess the evolution of colour in relation to anthocyanin profiles in different cherry cultivars, and notably to unravel any differences in the anthocyanin–colour relationships among different cherry cultivars. For this reason the number of samples taken from each cultivar is not sufficient to build a valid mathematical function to robustly predict the colour evolution during storage of each different cultivar. Thus, for a more comprehensive mathematical function describing the colour development in individual cherry cultivars, collection of more data from a large number of samples is recommended. The next step in such studies will be to predict the colour evolution and select the optimal storage mode for individual cherry cultivars.

When lumping the data for all four cultivars, all the negative correlations obtained statistical significance. The chromatic parameters L^*, a^*, b^*, chroma and hue angle cor-

Table 1
CIE 1976 ($L^*a^*b^*$) colour space (CIELAB) of cherry cultivars at two ripeness stages (ripe and partially-ripe) and influenced by storage at 15 ± 5 °C (room T) and 1.5 ± 0.5 °C (cool T) for year 2001

Cultivar	Storage	Chromatic coordinates			Chroma C^*_{ab}	Hue angle h_{ab}
		L^*	a^*	b^*		
Burlat						
P-ripe	Day 0	51.80 ± 7.09j	36.43 ± 6.76def	21.17 ± 2.34hi	42.44 ± 4.91e	30.95 ± 7.91j
P-ripe	Cool T day 30	42.23 ± 6.09h	37.00 ± 3.38efg	15.64 ± 2.50f	40.22 ± 3.71d	22.85 ± 2.86de
Ripe	Day 0	37.34 ± 4.75de	36.15 ± 5.89def	15.45 ± 4.41f	39.36 ± 7.09d	22.64 ± 3.06de
Ripe	Room T day 6	31.41 ± 3.48a	25.76 ± 7.85a	9.16 ± 4.74bc	27.48 ± 8.74a	18.70 ± 6.06abc
Ripe	Cool T day 30	30.36 ± 2.64a	25.38 ± 6.46a	7.78 ± 3.40a	26.59 ± 7.15a	16.32 ± 3.15a
Saco						
P-ripe	Day 0	52.24 ± 6.84j	35.84 ± 10.30de	29.43 ± 4.92j	47.57 ± 4.07f	40.48 ± 13.47k
P-ripe	Cool T day 30	41.25 ± 5.59gh	41.91 ± 3.55i	20.05 ± 4.50h	46.60 ± 4.44f	25.31 ± 4.62fg
Ripe	Day 0	38.52 ± 2.66ef	42.00 ± 2.11i	20.39 ± 3.16h	47.57 ± 4.07f	25.75 ± 2.85fgh
Ripe	Room T day 6	30.20 ± 1.34a	31.25 ± 3.21b	9.57 ± 2.08c	32.70 ± 3.66b	16.84 ± 1.93ab
Ripe	Cool T day 30	33.23 ± 3.09b	34.75 ± 5.76cde	12.32 ± 3.90d	36.92 ± 6.68c	18.98 ± 3.15bc
Summit						
P-ripe	Day 0	55.27 ± 7.10k	34.19 ± 10.23cd	29.20 ± 3.53j	45.98 ± 4.77f	41.76 ± 12.97k
P-ripe	Cool T day 30	46.43 ± 5.75i	38.34 ± 4.51fg	20.77 ± 2.58hi	43.82 ± 2.83e	28.76 ± 6.00ij
Ripe	Day 0	41.07 ± 5.29gh	38.54 ± 3.01fg	20.40 ± 4.13h	43.73 ± 3.83e	27.71 ± 4.41ghi
Ripe	Room T day 6	35.39 ± 2.05c	37.01 ± 3.30efg	16.19 ± 2.87f	40.43 ± 4.10d	23.44 ± 2.19ef
Ripe	Cool T day 30	35.72 ± 2.69cd	34.37 ± 2.56cd	14.08 ± 2.02e	37.17 ± 3.01c	22.18 ± 1.97de
Van						
P-ripe	Day 0	54.10 ± 8.43k	32.84 ± 11.34bc	31.02 ± 4.30k	46.52 ± 4.75f	44.79 ± 14.52e
P-ripe	Cool T day 30	39.42 ± 5.21fg	38.87 ± 3.31gh	18.68 ± 4.29g	43.26 ± 4.15e	25.41 ± 4.68fg
Ripe	Day 0	41.56 ± 4.15h	40.88 ± 2.30hi	21.81 ± 2.96i	46.41 ± 2.74f	28.01 ± 3.21hi
Ripe	Room T day 6	29.93 ± 1.38a	26.49 ± 4.40a	8.11 ± 2.27ab	27.73 ± 4.84a	16.73 ± 2.19ab
Ripe	Cool T day 30	33.17 ± 2.61b	34.08 ± 3.88cd	12.85 ± 3.03de	36.45 ± 4.68c	20.37 ± 2.41cd

Values are means ± SD ($n = 60$).
Means flanked by the same letter are not significantly different at $P < 0.05$ (Duncan's test).

related negatively ($P < 0.001$) with the total anthocyanins levels, but not with total phenols ($P > 0.05$) (Table 4). In all cases, the lowest values of chroma and hue angle corresponded to the samples having the highest anthocyanins content. It seems logic that yellowness (b^*) and lightness (L^*), and consequently hue and chroma values may correlate negatively with the anthocyanins levels, but it is more complex to understand why an increase in pigments causing redness gives lower redness value readings, i.e. decreased a^* values. This phenomenon was investigated and discussed in an early study on dark coloured fruit beverages by Eagerman, Clydesdale, and Francis (1973). They clearly demonstrated the presence of an ''inversion area'', where the increase anthocyanin pigment concentration, cyanidin-3-glucoside, where both the L^*, a^* and b^* values failed to correlate as expected to increases in the pigment concentration. In brief, this phenomenon is presumed to occur when increased pigment concentration both darkens the sample (e.g. the fruit or the fruit beverage) and increases the chroma. When this occurs, the colour scales are no longer tied linearly to the luminous transmittance (Eagerman et al., 1973). In our present work, the anthocyanins did indeed darken the cherries as their concentration increased, and therefore the chromaticity responses were no longer linear, and might, in fact, have reverted to correlate negatively. The darker the cherries, the more negative the correlation to anthocyanins levels. According to Little (1975) and McGuire (1992), hue angle and chroma give more information about spatial distribution of colours. Indeed, better correlation between these parameters and pigment concentrations have been obtained than when pigment concentrations were compared directly with the values from the colorimeter (McGuire, 1992).

For each relationship between total anthocyanins levels and chromatic parameters of the four sweet cherry cultivars, the regressions with highest determination coefficients and with significant ($P < 0.05$) regression coefficients were selected (Fig. 3). The best fit-adjustment (lower scattering) was found in the samples having less than 100 mg/100 g of fw of total anthocyanins.. This evidence suggested that the measure of chromatic parameters could be a good tool to predict the levels of anthocyanins in storage cherries and predict the beneficial human health effects of each sweet cherry cultivar, since anthocyanins exert antioxidant activity (Lapidot, Harel, Akiri, Granit, & Kanner, 1999; Matsumoto, Nakamura, Hirayama, Yoshiki, & Okubo, 2002; Wang et al., 1999). Therefore, for cherries for human consumption, it seems important to have a simple and non-destructive technique for anthocyanins content determination, and in this way easily and quickly assess and monitor cherry quality on a large number of cherries.

Table 2
CIE 1976 ($L^*a^*b^*$) colour space (CIELAB) of cherry cultivars at two ripeness stages (ripe and partially-ripe) and influenced by storage at 15 ± 5 °C (room T) and 1.5 ± 0.5 °C (cool T) for year 2002

Cultivar	Storage	Chromatic coordinates			Chroma C^*_{ab}	Hue angle h_{ab}
		L^*	a^*	b^*		
Burlat						
P-ripe	Day 0	45.05 ± 5.33h	43.17 ± 4.80l	23.37 ± 2.29i	49.15 ± 4.66l	28.53 ± 2.81i
P-ripe	Cool T day 30	38.13 ± 4.43g	36.08 ± 3.29j	15.97 ± 2.70g	39.49 ± 3.97k	23.72 ± 2.29gh
Ripe	Day 0	27.84 ± 2.35cd	18.50 ± 6.50e	4.98 ± 2.89bc	19.19 ± 7.03e	14.06 ± 3.17bcde
Ripe	Room T day 6	26.47 ± 0.67bc	9.31 ± 3.16ab	1.98 ± 0.82a	9.52 ± 3.25ab	11.83 ± 1.94abc
Ripe	Cool T day 30	24.43 ± 1.33a	9.76 ± 4.11ab	2.44 ± 1.25a	11.43 ± 11.04b	15.07 ± 9.88bcd
Saco						
P-ripe	Day 0	46.94 ± 8.14i	36.27 ± 7.80j	24.73 ± 6.15j	44.86 ± 3.38l	34.76 ± 12.51k
P-ripe	Cool T day 30	29.80 ± 2.94de	32.08 ± 4.89i	10.74 ± 3.47e	33.87 ± 5.74i	18.05 ± 2.91f
Ripe	Day 0	30.53 ± 2.73e	26.59 ± 6.56h	8.23 ± 3.71d	27.90 ± 7.31h	16.47 ± 3.67def
Ripe	Room T day 6	26.05 ± 0.66abc	12.16 ± 2.94cd	2.21 ± 0.79a	12.36 ± 3.03cd	10.10 ± 1.32a
Ripe	Cool T day 30	25.40 ± 1.24ab	13.57 ± 3.91d	2.82 ± 1.36a	13.87 ± 4.11d	11.25 ± 2.19ab
Summit						
P-ripe	Day 0	46.33 ± 5.43hi	41.02 ± 4.73k	25.72 ± 3.09j	48.64 ± 3.10e	32.30 ± 6.05jk
P-ripe	Cool T day 30	38.95 ± 4.25g	36.37 ± 2.41j	17.48 ± 2.90h	40.41 ± 3.01k	25.54 ± 3.30h
Ripe	Day 0	34.44 ± 3.55f	33.53 ± 4.55i	13.87 ± 4.10f	36.36 ± 5.65j	21.95 ± 3.87g
Ripe	Room T day 6	27.99 ± 1.59cd	17.77 ± 3.65e	4.40 ± 1.49b	18.32 ± 3.88e	13.63 ± 2.28bcd
Ripe	Cool T day 30	27.93 ± 2.55cd	23.72 ± 5.48g	7.47 ± 2.72d	24.90 ± 6.01g	16.92 ± 2.85ef
Van						
P-ripe	Day 0	59.14 ± 8.41j	23.36 ± 12.90fg	30.14 ± 5.37k	40.34 ± 4.31k	53.83 ± 19.15l
P-ripe	Cool T day 30	39.35 ± 14.30g	25.79 ± 8.95h	16.65 ± 10.51gh	33.09 ± 5.95i	31.52 ± 21.57j
Ripe	Day 0	28.64 ± 1.69d	21.53 ± 5.41f	5.87 ± 2.54c	22.34 ± 5.88f	14.57 ± 2.83cde
Ripe	Room T day 6	26.19 ± 0.62abc	7.79 ± 2.53a	1.42 ± 0.48a	7.93 ± 2.57a	10.50 ± 1.92a
Ripe	Cool T day 30	26.12 ± 1.04abc	10.71 ± 3.52bc	2.33 ± 0.90a	10.96 ± 3.62bc	12.17 ± 1.81abc

Values are means ± SD ($n = 60$).
Means flanked by the same letter are not significantly different at $P < 0.05$ (Duncan's test).

Table 3
Highlighting the different correlations between total anthocyanins and evolution of color parameters (L^*, a^*, b^*) chroma (C^*_{ab}) and hue angle (h_{ab}) of ripe cherries during storage among cultivars

	L^*	a^*	b^*	C^*_{ab}	h_{ab}
Total anthocyanins					
Burlat	−0.739	−0.878*	−0.819*	−0.871*	−0.843*
Saco	−0.702	−0.685	−0.734	−0.699	−0.768
Summit	−0.675	−0.721	−0.733	−0.729	−0.774
Van	−0.665	−0.839*	−0.727	−0.823*	−0.744
Sum anthocyanins all cultivars	−0.675**	−0.791***	−0.729***	−0.784***	−0.745***

* Indicates $P < 0.05$.
** Indicates $P < 0.01$.
*** Indicates $P < 0.001$ by Fisher's test.

Table 4
Correlation matrix between anthocyanins (total and individual anthocyanins) and total phenols with chromatic coordinates (L^*, a^*, b^*), chroma (C^*_{ab}) and hue angle (h_{ab}) of the four cherry cultivars during storage

	L^*	a^*	b^*	C^*_{ab}	h_{ab}
Cyanidin-3-glucoside	−0.548***	−0.709***	−0.629***	−0.729***	−0.548***
Cyanidin-3-rutinoside	−0.710***	−0.779***	−0.772***	−0.831***	−0.702***
Peonidin-3-glucoside	−0.449**	−0.660***	−0.529***	−0.657***	−0.435**
Peonidin-3-rutinoside	−0.533***	−0.732***	−0.624***	−0.742***	−0.539***
Pelargonidin-3-rutinoside	−0.684***	−0.724***	−0.735***	−0.776***	−0.667***
Total anthocyanins	−0.697***	−0.797***	−0.768***	−0.842***	−0.692***
Total phenols	−0.533***	−0.408**	−0.506***	−0.475**	−0.520***

* Indicates $P < 0.05$.
** Indicates $P < 0.01$.
*** Indicates $P < 0.001$ by Fisher's test.

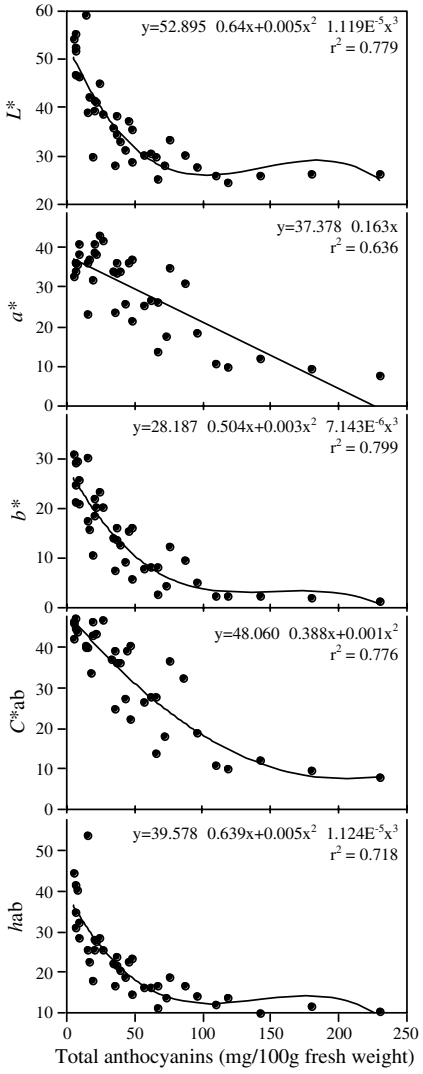

Fig. 3. Relationships between total anthocyanins and chromatic parameters of the four sweet cherry cultivar ($n = 39$). All the regression coefficients were significant ($P < 0.05$).

4. Conclusions

The chromatic parameters L^*, a^*, b^*, chroma and hue angle correlated negatively with the total anthocyanins levels, but not with the total phenols. In stored cherries the evolution of colour is a direct result of an increase in the levels of anthocyanins, particularly in the dominant anthocyanins cyanidin-3-rutinoside and -glucoside. Despite the variation on the anthocyanin levels in different cherry cultivars, which induces some variation in the degree of correlation between total anthocyanins and colour parameters, it is clear that colour measurements provide an easy assessment of the relative levels and changes of anthocyanins in different cherry cultivars during storage – and *vice versa*.

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